the fused cells. The fused cells can then be allowed to grow for approximately eight days. Supernatants from resultant hybridomas are collected and added to a plate that is first coated with goat anti-mouse Ig. Following washes, a label, such as, ^{125}I- pine tree polypeptides is added to each well followed by incubation. Positive wells can be subsequently detected by autoradiography. Positive clones can be grown in bulk culture and supernatants are subsequently purified over a Protein A column (Pharmacia).

[0101] Monoclonal antibodies and specific-binding fragments of the invention can be produced using alternative techniques, such as those described by Alting-Mees et al., "Monoclonal Antibody Expression Libraries: A Rapid Alternative to Hybridomas", *Strategies in Molecular Biology* 3:1-9 (1990), which is incorporated herein by reference. Similarly, binding partners can be constructed using recombinant DNA techniques to incorporate the variable regions of a gene that encodes a specific binding antibody. Such a technique is described in Larrick et al., *Biotechnology*, 7:394 (1989).

[0102] It is understood of course that many techniques could be used to generate antibodies against the polypeptides of the invention and that the above embodiments in no way limits the scope of the invention.

Nucleotides, Proteins, Antibodies, and Binding Proteins As Probes and Reagents

[0103] The disclosed nucleic acids, polypeptides, and antibodies directed against the disclosed polypeptides can be used in a variety of research protocols, such as in DNA arrays or as reagents. A sample of such research protocols are given in Sambrook et al. *Molecular Cloning: A Laboratory Manual*, 2 ed. Vol. 1-3, Cold Spring Harbor Laboratory Press, (1989), incorporated by reference. For example, the compiled

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sequences, polypeptides, etc., can serve as markers for cell specific or tissue specific expression of RNA or proteins. Similarly, this system can be used to investigate constitutive and transient expression of the genes encoding the cDNAs of SEQ ID NOS: 1-327 and the proteins encoded by these genes.

[0104] Further, the disclosed cDNA sequences can be used to determine the chromosomal location of the genomic DNA and to map genes in relation to this chromosomal location. The disclosed nucleotide sequence can be further used to identify additional genes related to the nucleotides of SEQ ID NOS: 1-334 and to establish evolutionary relatedness among species based on the comparison of sequences. The disclosed nucleotide and polypeptide sequences can be used to select for those genes or proteins that are homologous to the disclosed cDNAs or polypeptides, using well-established positive screening procedures such as Southern blotting and immunoblotting and negative screening procedures such as subtractive hybridization.

Method for Using Nucleic Acid Probes or Antibodies to Stage Embryos

[0105] Accurate staging of tree embryos is critical. It is known that different stages of tree embryos have different capacities as subjects for genetic transformation and genetic engineering. In addition, environmental requirements exhibited by embryos vary due to increasing physiologic age. Currently, the staging of tree embryogenesis is most accurately performed by an expert in the field who is very familiar with the morphological appearance of embryos at different stages. The cDNAs and related molecules of this invention can be used as markers for different stages of tree embryogenesis, thereby eliminating the need for a subjective eye to assess maturity

and potentially allowing for more accurate staging of tree embryos. Moreover, by monitoring the expression of the underlying genes, it is possible to determine when an embryo has reached a certain level of development even if that level does not correspond to a visible difference in embryo morphology. The relational database of this invention aids the ability to monitor expression levels and tailor research approaches, such as the use of DNA arrays, to the specific needs of the objective, i.e., staging.

[0106] The information provided in this invention can be used in whole or in part to stage embryos. For example, one or a multiplicity of nucleic acid molecules from SEQ ID NOS: 1-327 having an expression profile consistent with a particular embryo stage can be used in this invention. A researcher may find it beneficial to use oligonucleotide probes or antibodies, for example, that specifically recognize proteins derived from genes expressed during middle embryonic stages, or that specifically monitor expression levels for embryos that have reached maturity associated with late developmental stages. A researcher can quickly determine that an embryo subset has progressed to or through an embryonic stage with the use of this invention and make appropriate changes in conditions if necessary, e.g. alter growth media or other environmental conditions.

Method for Monitoring, Enhancing, or Determining Expression of Stage-Specific Genes

[0107] Expression patterns of SEQ ID NOS: 1-327 indicate that gene activation can be classified as stage-specific, such as in the case of SEQ ID NO: 327, otherwise known as "LP2-3." The promoter that drives such a gene can perform valuable functions. For example, a promoter from LP2-3 operatively linked to a reporter gene presented within an embryo system is expected to produce the reporter product under

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